

SEQUENCE LISTING

<110> Institut de Recherches Cliniques de Montreal SEIDAH, Nabil CHRÉTIEN, Michel MARCINKIEWICZ, Mieczyslaw LAAKSONEN, Reijo DAVIGNON, Jean

<120> MAMMALIAN SUBTILISIN/KEXIN ISOZYME SKI-1: A PROPROTEIN CONVERTASE WITH A UNIQUE CLEAVAGE SPECIFICITY

<130> IRCM

<140> PCT/CA99/01058

<141> 1999-11-04

<150> CA 2,249,648

<151> 1998-11-04

<160> 76

<170> PatentIn Ver. 2.1

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- Tyr Pro Ser Asp Phe Glu Val Ile Gln Ile Lys Glu Lys Gln Lys Ala 100 105 110
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- Pro Leu Lys Arg Ala Ser Leu Ser Leu Gly Ser Gly Phe Trp His Ala 165 170 175
- Thr Gly Arg His Ser Ser Arg Arg Leu Leu Arg Ala Ile Pro Arg Gln
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- Gly Ala Asn Val Arg Val Ala Val Phe Asp Thr Gly Leu Ser Glu Lys 210 215 220
- His Pro His Phe Lys Asn Val Lys Glu Arg Thr Asn Trp Thr Asn Glu 225 230 235 240
- Arg Thr Leu Asp Asp Gly Leu Gly His Gly Thr Phe Val Ala Gly Val 245 250 255
- Ile Ala Ser Met Arg Glu Cys Gln Gly Phe Ala Pro Asp Ala Glu Leu 260 265 270
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- Lys Val Trp Glu Leu Thr Ala Asn Asn Val Ile Met Val Ser Ala Ile 325 330 335
- Gly Asn Asp Gly Pro Leu Tyr Gly Thr Leu Asn Asn Pro Ala Asp Gln 340 345 350
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- Arg Phe Ser Ser Arg Gly Met Thr Thr Trp Glu Leu Pro Gly Gly Tyr 370 375 380
- Gly Arg Val Lys Pro Asp Ile Val Thr Tyr Gly Ala Gly Val Arg Gly 385 390 395 400
- Ser Gly Val Lys Gly Gly Cys Arg Ala Leu Ser Gly Thr Ser Val Ala 405 410 415
- Ser Pro Val Val Ala Gly Ala Val Thr Leu Leu Val Ser Thr Val Gln 420 425 430
- Lys Arg Glu Leu Val Asn Pro Ala Ser Val Lys Gln Ala Leu Ile Ala 435 440 445
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- Lys Leu Asp Leu Leu Arg Ala Tyr Gln Ile Leu Ser Ser Tyr Lys Pro 465 470 475 480
- Gln Ala Ser Leu Ser Pro Ser Tyr Ile Asp Leu Thr Glu Cys Pro Tyr 485 490 495
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- Ile Val Asn Val Thr Ile Leu Asn Gly Met Gly Val Thr Gly Arg Ile 515 520 525
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- Gln Glu Thr Ala Val Val Asp Asn Val Pro Ile Leu Gly Leu Tyr Gln 820 825 830
- Ile Pro Ala Glu Gly Gly Arg Ile Val Leu Tyr Gly Asp Ser Asn 835 840 845
- Cys Leu Asp Asp Ser His Arg Gln Lys Asp Cys Phe Trp Leu Leu Asp 850 855 860
- Ala Leu Leu Gln Tyr Thr Ser Tyr Gly Val Thr Pro Pro Ser Leu Ser 865 870 875 880
- His Ser Gly Asn Arg Gln Arg Pro Pro Ser Gly Ala Gly Leu Ala Pro 885 890 895
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- Gly Ala Met Val Ala Leu Ala Phe Phe Val Val Gln Ile Ser Lys Ala 1010 1015 1020
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Thr Ala Lys Ala Arg Asn Ser Phe Ile Ser Ser Ala Leu Lys Ser Ser 65 70 75 80

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Tyr Pro Ser Asp Phe Glu Val Ile Gln Ile Lys Glu Lys Gln Lys Ala 100 105 110

Gly Leu Leu Thr Leu Glu Asp His Pro Asn Ile Lys Arg Val Thr Pro 115 120 125

Gln Arg Lys Val Phe Arg Ser Leu Lys Phe Ala Glu Ser Asn Pro Ile 130 135 140

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- Ser Pro Val Val Ala Gly Ala Val Thr Leu Leu Val Ser Thr Val Gln 420 425 430
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- Ile Lys Val Lys Ile Ile Pro Thr Pro Pro Arg Ser Lys Arg Val Leu 610 615 620
- Trp Asp Gln Tyr His Asn Leu Arg Tyr Pro Pro Gly Tyr Phe Pro Arg 625 630 635 640
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- Glu Ala His Leu Gly Asp Pro Lys Pro Arg Pro Leu Pro Ala Cys Pro 915 920 925

His Leu Ser Trp Ala Lys Pro Gln Pro Leu Asn Glu Thr Ala Pro Ser 930 935 940

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Pro Gly Glu Ser Gly Ala Trp Asp Ile Pro Gly Gly Ile Met Pro Gly 980 985 990

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			ccc Pro													916
			aca Thr													964
			cgt Arg													1012

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			_			_					cgg Arg 840		-	-		3028
											cag Gln					3076
										-	tat Tyr				-	3124
											cct Pro					3172
		_				Arg	_	-			cat His					3220
	-	-	-		-		-		-		aaa Lys 920					3268
							-		_		cag Gln					3316

acg gcg ccc agt aac ctt tgg aaa cat cag aag cta ctc tcc att gac 336 Thr Ala Pro Ser Asn Leu Trp Lys His Gln Lys Leu Leu Ser Ile Asp 945 950 955	64
ctg gac aag gtg gtg tta ccc aac ttt cga tcg aat cgc cct caa gtg 341 Leu Asp Lys Val Val Leu Pro Asn Phe Arg Ser Asn Arg Pro Gln Val 960 965 970	12
agg ccc ttg tcc cct gga gag agc ggc gcc tgg gac att cct gga ggg 346 Arg Pro Leu Ser Pro Gly Glu Ser Gly Ala Trp Asp Ile Pro Gly Gly 975 980 985	60
atc atg cct ggc cgc tac aac cag gag gtg ggc cag acc att cct gtc 350 Ile Met Pro Gly Arg Tyr Asn Gln Glu Val Gly Gln Thr Ile Pro Val 990 995 1000	80
ttt gcc ttc ctg gga gcc atg gtg gtc ctg gcc ttc ttt gtg gta caa 355 Phe Ala Phe Leu Gly Ala Met Val Val Leu Ala Phe Phe Val Val Gln 1005 1010 1015 1020	56
atc aac aag gcc aag agc agg ccg aag cgg agg a)4
cgc ccg cag ctc atg cag cag gtt cac ccg cca aag acc cct tcg gtg 365 Arg Pro Gln Leu Met Gln Gln Val His Pro Pro Lys Thr Pro Ser Val 1040 1045 1050	52
tgaccggcag cctggctgac cgtgagggcc agagagagcc ttcacggacg gcgctggtgg 371	L2
gtgagccgag ctgtggtggc ggctggttta aaagggatcc agtttccagc tgcaggtttg 377	72
ttagagtetg ttetacatgg geetgeeete etgtgatggg cagaggetee tggtacateg 383	32
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ccgccggcag ctgtgcgcac caaagactcg ggagaactgg aaaggctgtc tggggtcttc 395	52
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acgcttgtta aaggctattt tctatattta ttgttgggaa aagtcacttt aaagacttgt 407	12
gctatttgga agcaaagcta ttttttttgt cagtggaatg cagttttttt actattccat 413	32
catgaggaac aacatagatt ccatgatctt tttaatgaca gtacagactg agatttgaag 419)2
gaaacatgca caaatctgta aaacatagac cttcgcttta tttttgtaag tatcacctgc 425	52

caccatgttt tgtaatttga ggtcttgatt tcaccattgt cggtgaagaa aattttcaat 4312
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<211> 1052

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<213> Homo sapiens

<400> 6

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20 25 30

Ala Pro Cys Pro Gly Cys Ser His Leu Thr Leu Lys Val Glu Phe Ser 35 40 45

Ser Thr Val Val Glu Tyr Glu Tyr Ile Val Ala Phe Asn Gly Tyr Phe 50 55 60

Thr Ala Lys Ala Arg Asn Ser Phe Ile Ser Ser Ala Leu Lys Ser Ser 65 70 75 80

Glu Val Asp Asn Trp Arg Ile Ile Pro Arg Asn Asn Pro Ser Ser Asp 85 90 95

Tyr Pro Ser Asp Phe Glu Val Ile Gln Ile Lys Glu Lys Gln Lys Ala 100 105 110

Gly Leu Leu Thr Leu Glu Asp His Pro Asn Ile Lys Arg Val Thr Pro 115 120 125

Gln Arg Lys Val Phe Arg Ser Leu Lys Tyr Ala Glu Ser Asp Pro Thr 130 135 140

Pro Leu Arg Arg Ala Ser Leu Ser Leu Gly Ser Gly Phe Trp His Ala 165 170 175

Thr Gly Arg His Ser Ser Arg Arg Leu Leu Arg Ala Ile Pro Arg Gln 180 185 190

Val Ala Gln Thr Leu Gln Ala Asp Val Leu Trp Gln Met Gly Tyr Thr

Gly Ala Asn Val Arg Val Ala Val Phe Asp Thr Gly Leu Ser Glu Lys His Pro His Phe Lys Asn Val Lys Glu Arg Thr Asn Trp Thr Asn Glu Arg Thr Leu Asp Asp Gly Leu Gly His Gly Thr Phe Val Ala Gly Val Ile Ala Ser Met Arg Glu Cys Gln Gly Phe Ala Pro Asp Ala Glu Leu His Ile Phe Arg Val Phe Thr Asn Asn Gln Val Ser Tyr Thr Ser Trp Phe Leu Asp Ala Phe Asn Tyr Ala Ile Leu Lys Lys Ile Asp Val Leu Asn Leu Ser Ile Gly Gly Pro Asp Phe Met Asp His Pro Phe Val Asp Lys Val Trp Glu Leu Thr Ala Asn Asn Val Ile Met Val Ser Ala Ile Gly Asn Asp Gly Pro Leu Tyr Gly Thr Leu Asn Asn Pro Ala Asp Gln Met Asp Val Ile Gly Val Gly Gly Ile Asp Phe Glu Asp Asn Ile Ala Arg Phe Ser Ser Arg Gly Met Thr Trp Glu Leu Pro Gly Gly Tyr Gly Arg Met Lys Pro Asp Ile Val Thr Tyr Gly Ala Gly Val Arg Gly Ser Gly Val Lys Gly Gly Cys Arg Ala Leu Ser Gly Thr Ser Val Ala Ser Pro Val Val Ala Gly Ala Val Thr Leu Leu Val Ser Thr Val Gln Lys Arg Glu Leu Val Asn Pro Ala Ser Met Lys Gln Ala Leu Ile Ala

Ser Ala Arg Arg Leu Pro Gly Val Asn Met Phe Glu Gln Gly His Gly

Lys Leu Asp Leu Leu Arg Ala Tyr Gln Ile Leu Asn Ser Tyr Lys Pro Gln Ala Ser Leu Ser Pro Ser Tyr Ile Asp Leu Thr Glu Cys Pro Tyr Met Trp Pro Tyr Cys Ser Gln Pro Ile Tyr Tyr Gly Gly Met Pro Thr Val Val Asn Val Thr Ile Leu Asn Gly Met Gly Val Thr Gly Arg Ile Val Asp Lys Pro Asp Trp Gln Pro Tyr Leu Pro Gln Asn Gly Asp Asn Ile Glu Val Ala Phe Ser Tyr Ser Ser Val Leu Trp Pro Trp Ser Gly Tyr Leu Ala Ile Ser Ile Ser Val Thr Lys Lys Ala Ala Ser Trp Glu Gly Ile Ala Gln Gly His Val Met Ile Thr Val Ala Ser Pro Ala Glu Thr Glu Ser Lys Asn Gly Ala Glu Gln Thr Ser Thr Val Lys Leu Pro Ile Lys Val Lys Ile Ile Pro Thr Pro Pro Arg Ser Lys Arg Val Leu Trp Asp Gln Tyr His Asn Leu Arg Tyr Pro Pro Gly Tyr Phe Pro Arg Asp Asn Leu Arg Met Lys Asn Asp Pro Leu Asp Trp Asn Gly Asp His Ile His Thr Asn Phe Arg Asp Met Tyr Gln His Leu Arg Ser Met Gly Tyr Phe Val Glu Val Leu Gly Ala Pro Phe Thr Cys Phe Asp Ala Ser Gln Tyr Gly Thr Leu Leu Met Val Asp Ser Glu Glu Glu Tyr Phe Pro Glu Glu Ile Ala Lys Leu Arg Arg Asp Val Asp Asn Gly Leu Ser Leu

- Val Ile Phe Ser Asp Trp Tyr Asn Thr Ser Val Met Arg Lys Val Lys
 725 730 735
- Phe Tyr Asp Glu Asn Thr Arg Gln Trp Trp Met Pro Asp Thr Gly Gly 740 745 750
- Ala Asn Ile Pro Ala Leu Asn Glu Leu Leu Ser Val Trp Asn Met Gly 755 760 765
- Phe Ser Asp Gly Leu Tyr Glu Gly Glu Phe Thr Leu Ala Asn His Asp 770 775 780
- Met Tyr Tyr Ala Ser Gly Cys Ser Ile Ala Lys Phe Pro Glu Asp Gly 785 790 795 800
- Val Val Ile Thr Gln Thr Phe Lys Asp Gln Gly Leu Glu Val Leu Lys 805 810 815
- Gln Glu Thr Ala Val Val Glu Asn Val Pro Ile Leu Gly Leu Tyr Gln 820 825 830
- Ile Pro Ala Glu Gly Gly Gly Arg Ile Val Leu Tyr Gly Asp Ser Asn 835 840 845
- Cys Leu Asp Asp Ser His Arg Gln Lys Asp Cys Phe Trp Leu Leu Asp 850 855 860
- Ala Leu Leu Gln Tyr Thr Ser Tyr Gly Val Thr Pro Pro Ser Leu Ser 865 870 875 880
- His Ser Gly Asn Arg Gln Arg Pro Pro Ser Gly Ala Gly Ser Val Thr 885 890 895
- Pro Glu Arg Met Glu Gly Asn His Leu His Arg Tyr Ser Lys Val Leu 900 905 910
- Glu Ala His Leu Gly Asp Pro Lys Pro Arg Pro Leu Pro Ala Cys Pro 915 920 925
- Arg Leu Ser Trp Ala Lys Pro Gln Pro Leu Asn Glu Thr Ala Pro Ser 930 935 940
- Asn Leu Trp Lys His Gln Lys Leu Leu Ser Ile Asp Leu Asp Lys Val 945 950 955 960
- Val Leu Pro Asn Phe Arg Ser Asn Arg Pro Gln Val Arg Pro Leu Ser

Pro Gly Glu Ser Gly Ala Trp Asp Ile Pro Gly Gly Ile Met Pro Gly 980 985 990

Arg Tyr Asn Gln Glu Val Gly Gln Thr Ile Pro Val Phe Ala Phe Leu 995 1000 1005

Gly Ala Met Val Val Leu Ala Phe Phe Val Val Gln Ile Asn Lys Ala 1010 1015 1020

Lys Ser Arg Pro Lys Arg Arg Lys Pro Arg Val Lys Arg Pro Gln Leu 025 1030 1035 1040

Met Gln Gln Val His Pro Pro Lys Thr Pro Ser Val 1045 1050

<210> 7

<211> 7

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Peptide

<220>

<221> MOD_RES

<222> (2)

<223> Xaa represents any amino acid.

<220>

<221> MOD RES

<222> (3)

<223> Xaa represents an alkyl or an aromatic hydrophobic amino acid.

<220>

<221> MOD_RES

<222> (4)..(6)

<223> Xaa represents any amino acid.

<220>

<221> MOD_RES

<222> (7)

<223> Xaa represents an acidic amino acid.

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Arg Xaa Xaa Xaa Xaa Xaa
<210> 8
<211> 7
<212> PRT
<213> Artificial Sequence
<220>
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<223> Xaa represents any amino acid.
<220>
<221> MOD RES
<222> (3)
<223> Xaa represents an alkyl or an aromatic hydrophobic
      amino acid.
<220>
<221> MOD RES
<222> (4)
<223> Xaa represents Lys, Leu, Phe or Thr.
<220>
<221> MOD RES
<222> (5)..(6)
<223> Xaa represents any amino acid.
<220>
<221> MOD_RES
<222> (7)
<223> Xaa represents an acidic amino acid.
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<210> 9
<211> 8
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<213> Artificial Sequence
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<220>
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<221> MOD_RES
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<223> Xaa represents an alkyl or an aromatic hydrophobic
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<223> Xaa represents any amino acid.
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<223> Xaa represents an acidic amino acid.
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 1
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<211> 8
<212> PRT
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<223> Xaa represents any amino acid.
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<223> Xaa represents an alkyl or an aromatic hydrophobic
      amino acid.
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<220>
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<222> (9)
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<221> MOD RES
<222> (3)
<223> Xaa represents an alkyl or an aromatic hydrophobic
      amino acid.
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<221> MOD RES
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<222> (5)..(8)
<223> Xaa represents any amino acid.
<220>
<221> MOD RES
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Arg Xaa Xaa Xaa Xaa Xaa Xaa
 1
                  5
<210> 13
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<211> 11

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<212> PRT
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<210> 14
<211> 13
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Peptide
<220>
<221> MOD_RES
<222> (1)
<223> Xaa represents orthoaminobenzoic acid.
<220>
<221> MOD_RES
<222> (13)
<223> Xaa represents 3-nitrotyrosine.
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Xaa Val Phe Arg Ser Leu Lys Tyr Ala Glu Ser Asp Xaa
<210> 15
<211> 23
<212> DNA
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<220>
<223> Description of Artificial Sequence:
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<222> (3)
<223> i
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<220>
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  <222> (9)
  <223> i
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  <221> modified_base
  <222> (12)
  <223> i
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 <222> (18)
 <223> i
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 <222> (21)
 <223> i
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 <210> 16
 <211> 31
 <212> DNA
 <213> Artificial Sequence
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 <222> (3)
 <223> i
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 <222> (6)
. <223> i
 <220>
 <221> modified_base
 <222> (9)
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<223> i

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 <223> i
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 <222> (15)
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. <222> (18)
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 <223> i
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 <222> (29)
 <223> i
 <400> 16
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                                                                     31
 <210> 17
 <211> 8
 <212> PRT
<213> Artificial Sequence
<220>
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<220>
<221> MOD_RES
<222> (5)
<223> Xaa represents histidine or phenylalanine.
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<220>

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<221> MOD_RES
<222> (6)
<223> Xaa represents valine or cysteine.
<400> 17
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<210> 18
<211> 11
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Peptide
<220>
<221> MOD_RES
<222> (4)
<223> Xaa represents valine or methionine.
<220>
<221> MOD RES
<222> (6)
<223> Xaa represents threonine or serine.
<220>
<221> MOD_RES
<222> (8)
<223> Xaa represents histidine or valine.
<220>
<221> MOD RES
<222> (10)
<223> Xaa represents alanine or threonine.
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Gly Thr Ser Xaa Ala Xaa Pro Xaa Val Xaa Gly
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                  5
<210> 19
<211> 28
<212> DNA
<213> Homo sapiens
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<400> 19

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<210>		
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<400>		
ctcga	gggct ctcagccgtg tgct	24
401.05		
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(213/	Altilitial bequence	
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	Oligonucleotide	
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gagga	agaga cagggataaa c	21
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	Oligonacieotide	
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gggata	atgct tagcattgac	20
<210>	23	
<211>	20	
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<223>	Description of Artificial Sequence: Oligonucleotide	
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< 400>	20	
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<220>		
	Description of Artificial Sequence:	
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<400>	29	
ggagco	atgg attgcacttt c	21
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<211> 27

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<210> 33 <211> 21						
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<210> 34 <211> 57 <212> DNA <213> Homo	sapiens					
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<210> 35 <211> 24 <212> DNA <213> Homo	sapiens					
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<213> Homo	sabreus					

<400> 36 ctcgagtgtc tgggcaacct ggcgcggg <210> 37 <211> 14 <212> PRT <213> Homo sapiens <400> 37 Lys Ala Gly Ser Arg Gly Leu Thr Ser Leu Ala Asp Thr Phe 5 10 <210> 38 <211> 27 <212> PRT <213> Homo sapiens <400> 38 Gly Gly Ala His Asp Ser Asp Gln His Pro His Ser Gly Ser Gly Arg 10 Ser Val Leu Ser Phe Glu Ser Gly Ser Gly Gly 20 <210> 39

28

<211> 18

<212> PRT

<213> Homo sapiens

<400> 39

Trp His Ala Thr Gly Arg His Ser Ser Arg Arg Leu Leu Arg Ala Ile 5 10

Pro Arg

<210> 40

<211> 17

<212> PRT

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<220>

<223> Description of Artificial Sequence: Peptide

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<210> 42
<211> 17
<212> PRT
<213> Homo sapiens
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Gly
<210> 43
<211> 15
<212> PRT
<213> Homo sapiens
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Arg Ala Ile Pro Arg Gln Val Ala Gln Thr Leu Gln Ala Asp Val
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<210> 44
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<212> PRT

<213> Homo sapiens

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Pro Gln Arg Lys Val Phe Arg Ser Leu
<210> 45
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<212> PRT
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Pro Gln Arg Lys Val Phe Arg Ser Leu Lys Tyr Ala Glu Ser Asp
<210> 46
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<222> (1)
<223> Xaa represents orthoaminobenzoic acid.
<220>
<221> MOD_RES
<222> (13)
<223> Xaa represents 3-nitrotyrosine.
<220>
<223> Description of Artificial Sequence: Peptide
<400> 46
Xaa Val Phe Arg Ser Leu Lys Tyr Ala Glu Ser Asp Xaa Ala
<210> 47
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<222> (1)
<223> Xaa represents orthoaminobenzoic acid.
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 <223> Xaa represent 3-nitrotyrosine.
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<212> PRT
<213> Rattus sp.
<400> 49
Lys Ala Gly Ser Arg Gly Leu Thr Thr Thr Ser Leu Ala Asp Thr Phe
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<210> 50
<211> 16
<212> PRT
<213> Homo sapiens
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Arg His Ser Ser Arg Arg Leu Leu Arg Ala Ile Pro Arg Gln Val Ala
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<211> 16

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Ala Ser Val Gly Arg Leu Ala Leu Ser Gln Glu Glu Pro Ala Pro Leu
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                                     10
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Asp Pro Arg Leu Arg Gln Phe Leu Gln Lys Ser Leu Ala Ala Ala Thr
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<212> PRT
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Leu Leu Lys Glu Leu Gln Asp Leu Ala Leu Gln Gly Ala Lys Glu Arg
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Met Ala Arg Ala Pro Gln Val Leu Phe Arg Gly Gly Lys Ser Gly Glu
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Glu Leu Glu Asn Leu Ala Ala Met Asp Leu Glu Leu Gln Lys Ile Ala
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<213> Bovis sp.
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Ala Ala Met Asp Leu Glu Leu Gln Lys Ile Ala Glu Lys Phe Ser Gly
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                                                           15
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<211> 16
<212> PRT
<213> Rattus sp.
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Ser Gln Thr Pro Leu Val Thr Leu Phe Lys Asn Ala Ile Ile Lys Asn

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